Notice of All wability	Application No. 09/640,882	Applicant(s) HALL, BARRY G.
	Examin r Kenneth R Horlick	Art Unit 1637

-- Th MAILING DATE of this communication app ars on the cover sheet with the corresp nd nce address--

All claims being allowable, PROSECUTION ON THE MERITS IS (OR REMAINS) CLOSED in this application. If not included h rewith (or previously mailed), a Notice of Allowance (PTOL-85) or other appropriate communication will be mailed in due course. **THIS NOTICE OF ALLOWABILITY IS NOT A GRANT OF PATENT RIGHTS.** This application is subject to withdrawal from issue at the initiative of the Office or upon petition by the applicant. See 37 CFR 1.313 and MPEP 1308.

1. ☒ This communication is responsive to *the response filed 10/20/03*.
2. ☒ Th allowed claim(s) is/are *1-10, 12, 13, and 15-43 (final claims 1-41)*.
3. ☒ Th drawings filed on *18 August 2000* are accepted by the Examiner.
4. ☐ Acknowledgment is made of a claim for foreign priority under 35 U.S.C. § 119(a)-(d) or (f).
   a) ☐ All b) ☐ Some* c) ☐ None of the:
      1. ☐ Certified copies of the priority documents have been received.
      2. ☐ Certified copies of the priority documents have been received in Application No. _____ .
      3. ☐ Copies of the certified copies of the priority documents have been received in this national stage application from the International Bureau (PCT Rule 17.2(a)).

   * Certified copies not received: _____.
5. ☒ Acknowledgment is made of a claim for domestic priority under 35 U.S.C. § 119(e) (to a provisional application).
   (a) ☐ The translation of the foreign language provisional application has been received.
6. ☐ Acknowledgment is made of a claim for domestic priority under 35 U.S.C. §§ 120 and/or 121.

Applicant has THREE MONTHS FROM THE "MAILING DATE" of this communication to file a reply complying with the requirements noted below. Failure to timely comply will result in ABANDONMENT of this application. **THIS THREE-MONTH PERIOD IS NOT EXTENDABLE.**

7. ☐ A SUBSTITUTE OATH OR DECLARATION must be submitted. Note the attached EXAMINER'S AMENDMENT or NOTICE OF INFORMAL PATENT APPLICATION (PTO-152) which gives reason(s) why the oath or declaration is deficient.

8. ☐ CORRECTED DRAWINGS must be submitted.
   (a) ☐ including changes required by the Notice of Draftsperson's Patent Drawing Review (PTO-948) attached
      1) ☐ hereto or 2) ☐ to Paper No. ____.
   (b) ☐ including changes required by the proposed drawing correction filed _____, which has been approved by the Examiner.
   (c) ☐ including changes required by the attached Examiner's Amendment / Comment or in the Office action of Paper No. _____.

Identifying indicia such as the application number (see 37 CFR 1.84(c)) should be written on the drawings in the front (not the back) of each sheet.

9. ☐ DEPOSIT OF and/or INFORMATION about the deposit of BIOLOGICAL MATERIAL must be submitted. Note the attached Examiner's comment regarding REQUIREMENT FOR THE DEPOSIT OF BIOLOGICAL MATERIAL.

Attachment(s)

1☐ Notice of References Cited (PTO-892)
2☐ Notice of Informal Patent Application (PTO-152)
3☐ Notice of Draftperson's Patent Drawing Review (PTO-948)
4☐ Interview Summary (PTO-413), Paper No.____ .
5☐ Information Disclosure Statements (PTO-1449), Paper No. ____.
6☒ Examiner's Amendment/Comment
7☐ Examiner's Comment Regarding Requirement for Deposit of Biological Material
8☐ Examiner's Statement of Reasons for Allowance
9☐ Other .

1. A request for continued examination under 37 CFR 1.114 was filed in this application after appeal to the Board of Patent Appeals and Interferences, but prior to a decision on the appeal. Since this application is eligible for continued examination under 37 CFR 1.114 and the fee set forth in 37 CFR 1.17(e) has been timely paid, the appeal has been withdrawn pursuant to 37 CFR 1.114 and prosecution in this application has been reopened pursuant to 37 CFR 1.114. Applicant's submission filed on 10/20/03 has been entered.

2. Any inquiry concerning this communication or earlier communications from the examiner should be directed to Kenneth R Horlick whose telephone number is 703-308-3905. The examiner can normally be reached on Monday-Thursday 6:30AM-5:00 PM.

If attempts to reach the examiner by telephone are unsuccessful, the examiner's supervisor, Gary Benzion can be reached on 703-308-1119. The fax phone number for the organization where this application or proceeding is assigned is (703) 872-9306.

Any inquiry of a general nature or relating to the status of this application or proceeding should be directed to the receptionist whose telephone number is 703-308-0196.

Kenneth R. Horlick, Ph. D.
Kenneth R Horlick
Primary Examiner
Art Unit 1637

10/27/03

ALLOWED CLAIMS/ TJ

1. (currently amended) A method of predicting the evolutionary potential of a mutant resistance gene comprising:

preparing a mutant resistance gene that confers a selectable resistance phenotype to a host cell, said preparing comprising successive rounds of mutagenesis and selection ~~until no further~~ where each of the rounds confers enhancement of the resistance phenotype until clinical resistance is achieved ~~is perceived~~, the mutant resistance gene either including two or more nucleic acid modifications or encoding a mutant polypeptide including two or more amino acid modifications; and

determining whether the mutant resistance gene is likely to evolve through two or more independent mutation events, where each independent mutation event confers an enhancement of the resistance phenotype.

2. (original) The method according to claim 1, wherein the mutant resistance gene encodes the mutant polypeptide.

3. (original) The method according to claim 2, wherein each of the two or more amino acid modifications is selected independently from the group consisting of additions, deletions, substitutions, duplications, and rearrangements.

4. (previously presented) The method according to claim 2, wherein said determining comprises:

identifying two or more mutations of the mutant resistance gene which affect the amino acid sequence of the mutant polypeptide;

preparing a first set of singly mutated resistance genes each of which encodes a singly mutated polypeptide consisting of one of the two or more amino acid modifications of the mutant polypeptide;

inserting each of the first set of singly mutated resistance genes individually into one of a first set of host cells; and

selecting one or more of the first set of singly mutated resistance genes which confer a selectable enhancement of the resistance phenotype to the host cells of the first set, wherein the absence of any selected singly mutated resistance genes indicates that the mutant resistance gene is unlikely to evolve through independent mutations.

5. (original) The method according claim 4, wherein said selecting comprises:

introducing the first set of host cells onto a selection media and

collecting individual host cells of the first set which grow on the selection media.

6. (original) The method according to claim 5, wherein the selection media comprises at least one antibiotic and the mutant resistance gene is an antibiotic resistance gene.

7. (original) The method according to claim 6, wherein the selection media comprises a plurality of antibiotics.

8. (original) The method according to claim 5, wherein the selection media comprises at least one antiviral agent and the mutant resistance gene is an antiviral resistance gene.

9. (original) The method according to claim 5, wherein the selection media comprises at least one antifungal agent and the mutant resistance gene is an antifungal resistance gene.

10. (original) The method according to claim 5, wherein the selection media comprises at least one antimalarial agent and the mutant resistance gene is an antiprotozoal resistance gene.

12. (previously presented) The method according to claim 4, further comprising:

modifying the selected singly mutated resistance genes, wherein said modifying comprises introducing an additional mutation in the selected singly mutated resistance gene to prepare one or more doubly mutated resistance genes each of which encodes a doubly mutated polypeptide;

inserting each of the one or more doubly mutated resistance genes individually into a second set of host cells; and

selecting one or more of the doubly mutated resistance genes which confers a selectable enhancement of the resistance phenotype to the host cells of the second set.

13. (previously presented) The method according claim 12, wherein said selecting one or more of the doubly mutated resistance genes comprises:

introducing the host cells of the second set onto a selection media and

collecting the host cells of the second set which grow on the selection media.

15. (currently amended) The method according to claim ~~14~~ 12, further comprising:

repeating the steps of modifying, inserting, selecting, and assessing in succession until each of the two or more mutations of the mutant resistance gene have been recreated, indicating that the mutant resistance gene is likely to evolve through independent mutations.

16. (original) The method according to claim 1, wherein the mutant resistance gene includes two or more nucleic acid modifications, the two or more nucleic acid modifications affecting expression levels of the encoded polypeptide.

17. (original) The method according to claim 16, wherein the two or more nucleic acid modifications are present in a promoter region of the mutant resistance gene.

18. (previously presented) The method according to claim 1, wherein said preparing comprises:

providing a resistance gene;

introducing a plurality of mutations into the resistance gene to produce a mutated resistance gene;

inserting the mutated resistance gene into a host cell;

selecting the host cell which exhibits the strongest resistance phenotype;

isolating the mutated resistance gene from the selected host cell; and

repeating at least one subsequent round of said providing, introducing, inserting, selecting, and isolating until the mutated resistance gene of a subsequent round exhibits no further enhancement of the resistance phenotype relative to the mutated resistance gene of a preceding round.

19. (original) The method according to claim 18, wherein said introducing is carried out by DNA shuffling, error-prone PCR, or cassette mutagenesis.

20. (original) The method according to claim 18, wherein said inserting is carried ut by

ligating the mutant resistance gene into a plasmid and

treating the host cell under conditions effective to incorporate the plasmid into the host cell.

21. (original) The method according to claim 20, wherein said treating is carried out by electroporation.

22. (presently presented) The method according to claim 20, wherein the plasmid is selected from the group consisting of pACSE, pACSE2, and pACSE3.

23. (original) The method according to claim 1, wherein the host cell is *Escherichia coli*.

24. (currently amended) A method of screening a drug for anti-pathogenic activity against a pathogen including a mutant anti-pathogenic resistance gene, the method comprising:

providing a host cell comprising a mutant anti-pathogenic resistance gene either including two or more nucleic acid modifications or encoding a mutant anti-pathogenic polypeptide which includes two or more amino acid modifications, wherein the mutant anti-pathogenic resistance gene or mutant anti-pathogenic polypeptide confers a selectable advantage to the host cell, the mutant anti-pathogenic resistance gene having been prepared by successive rounds of mutagenesis and selection ~~until no further~~ where each of the rounds confers enhancement of the resistance phenotype until clinical resistance is achieved is ~~perceived~~ and having been demonstrated to be likely to evolve through two or more independent mutation events, where each independent mutation event confers an enhancement of the resistance phenotype;

growing the host cell on a selection media comprising a candidate drug or combinations thereof; and

determining whether the host cell is capable of growing on the selection media, wherein absence of host cell growth and/or proliferation indicates anti-pathogenic activity for the candidate drug or combinations thereof.

25. (previously presented) The meth d according to claim 24, wherein said providing the host cell comprises:

providing an anti-pathogenic resistance gene;

introducing a plurality of mutations into the anti-pathogenic resistance gene to produce a mutated anti-pathogenic resistance gene;

inserting the mutated anti-pathogenic resistance gene into a host cell;

selecting the host cell which exhibits the strongest resistance phenotype;

isolating the mutated resistance gene from the selected host cell;

repeating said providing, introducing, inserting, selecting, and isolating until the mutated resistance gene of a subsequent round exhibits no further enhancement of the resistance phenotype relative to the mutated resistance gene of a preceding round; and

determining whether the mutant resistance gene is likely to evolve through two or more independent mutation events, where each independent mutation event confers an enhancement of the resistance phenotype.

26. (original) The method according to claim 25, wherein the mutant anti-pathogenic resistance gene includes two or more nucleic acid modifications.

27. (original) The method according to claim 26, wherein the two or more nucleic acid modifications are present in a promoter region.

28. (original) The method according to claim 25, wherein the mutant anti-pathogenic resistance gene encodes the mutant anti-pathogenic polypeptide.

29. (original) The method according to claim 25, wherein said introducing is carried out by DNA shuffling, error-prone PCR, or cassette mutagenesis.

30. (previously presented) The method according to claim 25, wherein said inserting is carried out by

ligating the mutated anti-pathogenic resistance gene into a plasmid and

treating the host cell under conditions effective to incorporate the plasmid into the host cell.

31. (original) The method according to claim 25, wherein said inserting is carried out by

ligating the mutant anti-pathogenic resistance gene into a plasmid and

treating the host cell under conditions effective to incorporate the plasmid into the host cell.

32. (previously presented) The method according to claim 30, wherein the plasmid is selected from the group consisting of pACSE, pACSE2, and pACSE3.

33. (original) The method according to claim 24, wherein the host cell is *Escherichia coli*.

34. (original) The method according to claim 24, wherein th anti-pathogenic resistance gene and mutant anti-pathogenic resistance genes are selected from th group consisting of (i) antibiotic resistance gene and mutant antibiotic resistance gene, (ii) anti-viral resistance gene and mutant anti-viral resistance gene, (iii) anti-fungal resistance gene and mutant anti-fungal resistance gene, and (iv) anti-protozoal resistance gene and mutant anti-protozoal resistance gene.

35. (original) A method of assessing the potential longevity of a candidate anti-pathogenic drug comprising:

providing a resistance gene encoding a polypeptide which is ineffective against a candidate anti-pathogenic drug;

introducing multiple mutations into the resistance gene to produce a mutant resistance gene which encodes a mutant polypeptide including two or more amino acid modifications, wherein the mutant polypeptide is effective against the candidate anti-pathogenic drug;

determining the minimum number of mutations required to overcome the activity of the candidate anti-pathogenic drug, wherein the greater the minimum number of mutations, the greater the potential longevity of the candidate anti-pathogenic drug.

36. (original) The method according to claim 35, wherein the resistance gene and mutant resistance gene are selected from the group consisting of (i) antibiotic resistance gene and mutant antibiotic resistance gene, (ii) anti-viral resistance gene and mutant anti-viral resistance gene, (iii) anti-fungal resistance gene and mutant anti-fungal resistance gene, and (iv) anti-protozoal resistance gene and mutant anti-protozoal resistance gene.

37. (original) The method according to claim 35, wherein said introducing is carried out by DNA shuffling, error-prone PCR, or cassette mutagenesis.

38. (original) The method according to claim 35 further comprising after said introducing:

inserting the mutant resistance gene into a host cell; and

assaying whether the host cell can survive when grown on a selection media comprising the candidate anti-pathogenic drug, wherein survival of the host cell indicates that the mutant polypeptide is effective against the candidate anti-pathogenic drug.

39. (original) The method according to claim 38, wherein said inserting is carried out by

ligating the mutant resistance gene into a plasmid and

treating the host cell under conditions effective to incorporate the plasmid into the host cell.

40. (original) The method according to claim 39, wherein said treating is carried out by electroporation.

41. (original) The method according to claim 35, wherein said determining comprises:

identifying the two or more amino acid modifications of the mutant polypeptide;

preparing a plurality of singly mutated resistance genes each of which encodes a singly mutated polypeptide, the singly mutated polypeptide consisting of one of the two or more amino acid modifications of the mutant polypeptide;

inserting each of the plurality of singly mutated resistance genes individually into a first set of host cells; and

assaying whether any of the first set of host cells can survive when grown on a selection media comprising the candidate anti-pathogenic drug, wherein survival of any of the first set of host cells indicates that a single mutational event is all that is required to overcome the efficacy of the candidate anti-pathogenic drug.

42. (original) The method according to claim 41, further comprising:

modifying th selected singly mutated resistance genes, wherein said modifying comprises introducing an additional mutation into each of the singly mutated resistance genes to prepare one or more doubly mutated resistance genes each of which encodes a doubly mutated polypeptide, the doubly mutated polypeptide consisting of two of the two or more modifications of the mutant polypeptide;

second inserting each of the one or more doubly mutated genes into a second set of host cells; and

assaying whether any of the second set of host cells can survive when grown on the selection media comprising the candidate anti-pathogenic drug, wherein survival of any of the host cells indicates that two mutational events are all that is required to overcome the efficacy of the candidate anti-pathogenic drug.

43. (original) The method according to claim 42, further comprising:

optionally repeating the steps of modifying, second inserting, and assaying in succession until each of the mutations of the mutant resistance gene have been recreated, indicating that each of the two or more amino acid modifications of the mutant polypeptide are required to overcome the efficacy of the candidate anti-pathogenic drug.